

Supplementary Material

Longitudinal biomarkers in amyotrophic lateral sclerosis

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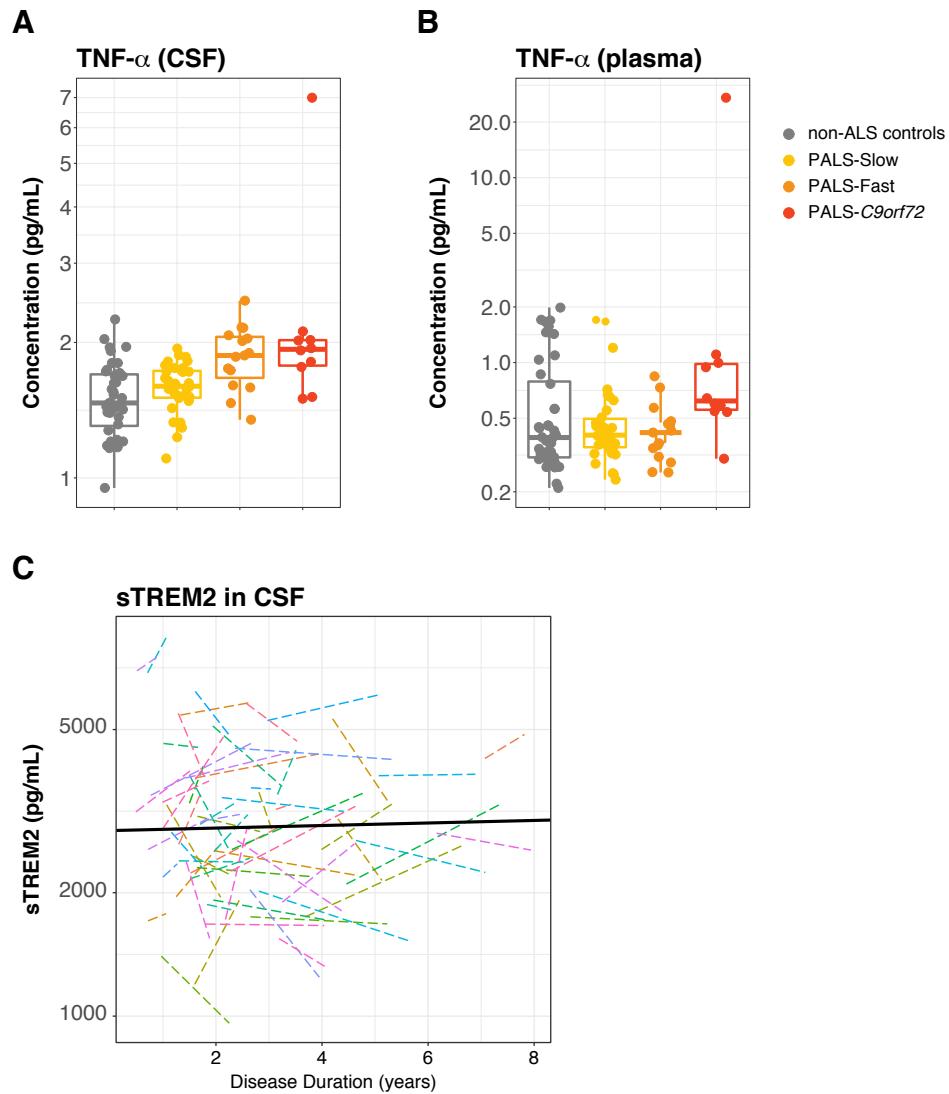
Supplementary Table 1. CSF cytokine comparison in PALS vs non-ALS control subjects.

Analyte	% relative to controls	p-value	adjusted p-value
Interleukin (IL)-18	149%	< 0.01	< 0.01
Monocyte chemoattractant protein (MCP)-1	129%	< 0.01	< 0.01
Macrophage inflammatory protein (MIP)-1 α	120%	< 0.01	0.01
B cell-attracting chemokine (BCA)-1	111%	0.59	0.73
Tumor necrosis factor (TNF)- α	111%	0.03	0.15
Soluble CD40 ligand (sCD40L)	111%	0.17	0.35
Platelet-derived growth factor (PDGF)-BB	110%	0.16	0.35
Interferon- γ -inducible protein (IP)-10	110%	0.48	0.68
IL-5	109%	0.13	0.31
IL-9	108%	0.32	0.57
I-309	108%	0.46	0.67
IL-10	107%	0.49	0.68
Granulocyte macrophage-colony stimulating factor (GM-CSF)	107%	0.04	0.15
IL-3	106%	0.31	0.57
IL-12 p70	106%	0.03	0.15
Macrophage-derived chemokine (MDC)	105%	0.08	0.23
IL-1 β	105%	0.11	0.26
IL-17A	105%	0.04	0.15
Granulocyte-colony stimulating factor (G-CSF)	105%	0.39	0.61
Vascular endothelial growth factor (VEGF)-A	105%	0.05	0.16
IL-13	104%	0.04	0.15
IL-1 α	104%	0.44	0.66
TNF- β	104%	0.02	0.14
Fms-related tyrosine kinase 3 ligand (Flt-3L)	104%	0.54	0.72
IFN- α 2	103%	0.26	0.50
IL-15	103%	0.41	0.63
Fibroblast growth factor (FGF)-2	103%	0.02	0.15
Epidermal growth factor (EGF)	103%	0.13	0.31
IL-7	103%	0.55	0.72
IL-12 p40	102%	0.50	0.68
Eotaxin-1	102%	0.17	0.35
IL-2	102%	0.37	0.61
Interferon (IFN)- γ	102%	0.56	0.73
Stromal cell-derived factor 1 (SDF-1) $\alpha+\beta$	101%	0.89	0.93
IL-16	101%	0.90	0.93
MCP-3	101%	0.58	0.73
Fractalkine	101%	0.86	0.92
MCP-2	100%	1.00	1.00

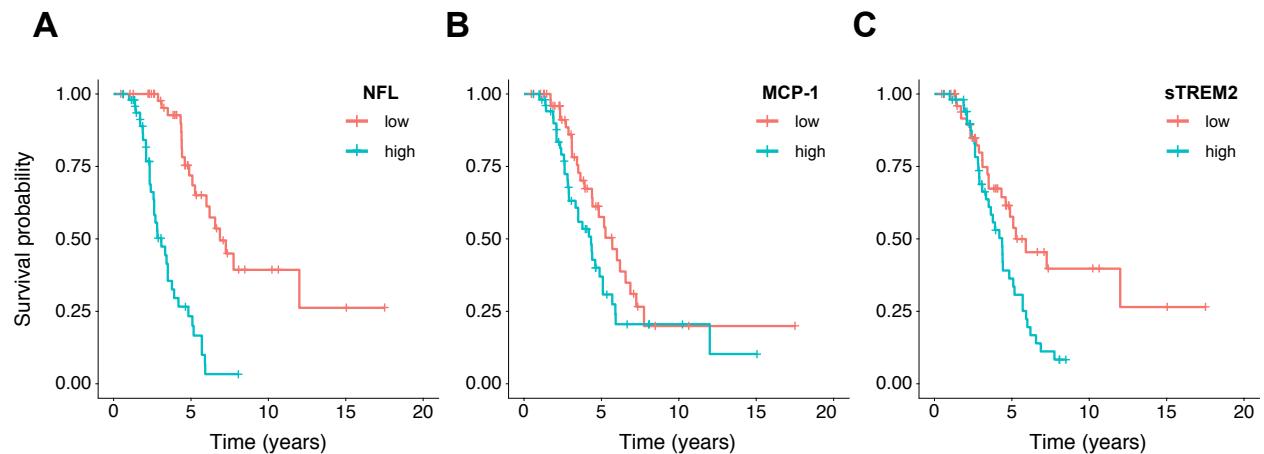
Analyte	% relative to controls	p-value	adjusted p-value
IL-1 receptor (IL-1R) α	100%	0.93	0.94
Growth-regulated oncogene (GRO) α	99%	0.81	0.89
IL-4	99%	0.79	0.88
MIP-1 β	99%	0.82	0.89
IL-6	97%	0.77	0.88
IL-8	97%	0.70	0.83
MCP-4	97%	0.61	0.74
Eotaxin	96%	0.76	0.88
IL-28A	96%	0.35	0.61
Epithelial neutrophil-activating protein (ENA)	95%	0.23	0.46
TNF-related apoptosis-inducing ligand (TRAIL)	93%	0.23	0.46
Regulated upon activation, normal T cell expressed and secreted (RANTES)	92%	0.36	0.61
Leukemia inhibitory factor (LIF)	92%	0.06	0.18
IL-33	92%	0.04	0.15
IL-20	91%	0.04	0.15
Transforming growth factor (TGF)- α	91%	0.04	0.15
IL-21	91%	0.10	0.25
Thyroid peroxidase (TPO)	91%	0.03	0.15
IL-23	89%	0.01	0.09
Thymus and activation-regulated chemokine (TARC)	88%	0.38	0.61
MIP-1 δ	86%	0.06	0.18
6CKine	85%	< 0.01	0.03
Stem cell factor (SCF)	85%	0.03	0.15
Cutaneous T-cell-attracting chemokine (CTACK)	74%	< 0.01	< 0.01
PDGF-AA	63%	< 0.01	< 0.01

For eotaxin-3 and thymic stromal lymphopoietin (TSLP), more than 90% of Luminex readouts were outside the range of quantification, and therefore excluded from further analyses. Only 1.5% of observations for remaining analytes were marked as out of range.

Supplementary Figure 1. TNF- α in (A) CSF and (B) plasma in disease progression subgroups. (C) Longitudinal sTREM2 in CSF from PALS.



Supplementary Figure 2. Summary of survival analyses in PALS with CSF NFL and cytokines.



Distinct curves representing cumulative survival in PALS with different CSF baseline levels. Estimated hazard ratios between “high” versus “low” analytes: (A) NFL: 9.99 (95% CI: 4.75-21.0), p < 0.001; (B) MCP-1: 1.36 (95% CI: 0.80 - 2.32), p = 0.260; (C) sTREM2: 1.36 (95% CI: 0.77-2.41), p = 0.295; N=102 with 54 observed deaths.